CLAIMS

1. A stable chloroplast transformation and expression vector which is capable of introducing multiple genes into a selected plant by a single integration event, wherein each step of said multiple genes is carried out by an enzyme encoding a heterologous DNA sequence which comprises an expression cassette, comprising as operably linked components, in the 5' to the 3' direction of translation, a promoter operative in said plastids which drives a multi-gene operon, a selectable marker sequence, the multi-gene operon which is functional to co-express multiple enzymes in the plastids, a transcription termination region functional in said plastids, and flanking each side of the expression cassette, flanking DNA sequences which are homomlogous to DNA sequences inclusive of a spacer sequence of the target plastid genome, whereby stable integration of the heterologous coding sequence into the chloroplast genome of the target plant is facilitated throughout homologous recombination of the flanking sequence with the homologous sequences in the target plastid gene.

2. A vector of claim 1, wherein a gene of the operon codes for an insecticidal toxin crystal protein.

3. A vector of claim 2, wherein the insecticidal toxin crystal protein is a *Bacillus thumngensis* (Bt) crystal protein.

4. A vector of claim 3, wherein another gene of the operon codes for another insecticidal crystal protein with a different mode of action

5. A vector of claim 4, wherein the multi-gene operon is functional to co-express, in addition to a Bt insecticidal toxin gene, a non-Bt insecticidal toxin gene selected from at least one of the group of cholesterol oxidase, alpha-amylate inhibitors, protease inhibitors, cowpea trypsin inhibitors and the potato proteinase inhibitor II, whereby the "pyramiding" of the toxin product tends to retard the ability of insects to adapt to the insecticidal effect of the transgenic target plants.

6. A vector of claim 4 or 5, wherein a second gene of the operon codes for a putative chaperonin which facilitates the folding of the Bt crystal toxin protein to form proteotically stable cuboidal crystals.

7. A vector of claim 3, wherein the operon includes one of the 133 genes shown in the article

MMBR, (September, 1998, pages 805-873, Vol 62, No. 4, Revision of the Nomenclature for the Bt-Pesticidal (insecticidal) Crystal Proteins by Crickmon, et al.), wherein at least one of the genes of the operon codes for a Bt insecticidal crystal protein and another gene codes for a putative corresponding chaperonin which facilitates the folding and the corresponding chaperonin facilitates the folding of the Bt protein to proteolytically stable cuboidal crystals shown in that MMBR article.

8. A vector of claim 1, wherein at least one of the gene of the operon codes for a biopharmaceutical protein.

9. A vector of claim 7, wherein another gene of the operon codes for a putative chaperonin which facilitates the folding of the protein.

10. A vector of claim 8, wherein the protein is insulin or human albumin.

11. A vector of claim 7, wherein another gene of the operon codes for another biopharmaceutical protein other than the gene which codes for the putative chaperonin, which protein is expressed in stoichimetric ratio.

12. A vector of claim 10, wherein the genes of the operon, other than the gene which codes for the putative chaperonin, codes for biopharmaceutical proteins which are expressed in stoichimetric ratio.

13. A vector of claims 7, 8, 9 or 10 which comprises collecting the protein product in a folded configuration, thereby enhancing their stability, and facilitating single step purification.

14. A method of combating insects which comprises applying to the insects or their habitat an insecticidally amount of the insecticidal crystal protein of claim 4.

15. A method of transforming a chloroplast of a selected plant species or the progeny thereof to confer insect resistance and producing on a large-scale foreign protein, said method comprising the steps of:

stably transforming the chloroplast of selected plant cells to express at least one insecticidal toxin protein and a chaperonin, growing the transformed plant cells under conditions which allow the expression of said insecticidal toxin protein and chaperonin.

16. The method of claim 14, further comprising the steps of culturing said plant cells in a plant growth medium comprising spectinomycin, and selecting transformed plant cells capable of growth in the presence of said spectinomycin.

17. The method of claim 15, further comprising regenerating a transformed plant from said transformed plant cells.

18. A transformed plant which has been transformed by the method of any one of claims 14-16.

19. The transformed plant of claim 18, wherein said plant contains a high accumulation of insecticidal toxin proteins in said plant's leaves, including mature and old bleached leaves.

20. The progeny, including seeds, of the transformed plant of claim 18.

21. A vector of claim 1, wherein the biosynthetic pathway is a bioremediation system that functions to degrade inorganic and organic metal compounds in contaminated sites.

22. A vector of claim 21, wherein the expression cassette does not contain a terminator.

23. A vector of claim 21 or claim 22, wherein the operon contains the mercury resistance coding sequences encoding enzymes Mer A and Mer B.

24. The vector of claim 23, wherein the bioremediation pathway is driven by a single promoter.

25. The chloroplast transformation and expression vector of claim 24, wherein enzymes of the bioremediation pathway are expressed in stoichiometric amounts.

26. A vector of claim 25, wherein the inorganic compounds are selected from at least one of the group consisting of divalent cations of mercury, nickel, cobalt, trivalent cations of gold, and monovalent cations of silver.

27. A vector of claim 25, wherein the organic compounds are selected from at least one of the group consisting of alkyl mercury, alkenyl mercury, alkynyl mercury, aromatic mercury compounds, alkyl lead compounds, alkyl arsenic compounds and alkyl cadmium compounds.

28. A method of transforming a chloroplast of a selected plant species or the progeny thereof to confer greater resistance to metal ions than the corresponding parental plant which does not require several back crosses to create complete pathway that detoxifies mercury and organiomercurial, said method comprising the steps of:

stably transforming the chloroplast of a plant by inserting an expression cassette containing the mercury resistance coding sequences of claim 21 into a plant species or the progeny

thereof, growing the transforming plant species under conditions which allow the expression of said expression cassette.

29. The method of claim 28, further comprising culturing said plant in a plant growth medium comprising a selector for the corresponding selectable marker of claim 1, and selecting transformed plant cells capable of growth in the presence of said selector.

30. The method of claim 29, further comprising regenerating a transformed plant from said transformed plant cells.

31. A stably transformed plant which has been transformed by the methods of any one of claims 28-30.

32. The progeny, including seeds, of the stably transformed plant of claim 31.

33. A method of phytoremediation of mercury and organomercurials in soil and ground water, said method comprising the steps of:

planting the stably transformed plant of either claim 31 or claim 32 in soil contaminated with mercury and organomercurials and allowing said plants to grow.

34. A method of phytoremidation which does not require several back crosses to create complete pathway that detoxifies mercury and organomercurial, said method comprising the methods of claims 28-30.

35. The plants of claim 33, wherein the plant contains products of the bioremediation pathway.

36. The products of the stably transformed plant of either claim 31 or claim 32, wherein said products are metals that are reduced by the enzymes of the bioremediation pathway.

37. The vector of claim 23 which is capable of introducing a multiple-step biosymethtic pathway into a selected photosynthetic cell by a single integration event

38. The vector of claim 37, wherein the biosynthetic pathway degrades inorganic and organic mercury compounds.

39. A vector of claim 38, wherein the bioremediation pathway is driven by a single promoter.

40. A vector of claim 38, wherein the enzymes of the bioremediation pathway are expressed in stoichiometric amounts.

41. A vector of claim 38, wherein the inorganic compounds are selected from at least one of

a group consisting of divalent cations of mercury, nickel, cobalt, trivalent cations of gold, and monovalent cations of silver.

42. A vector of claim 38, wherein the organic compounds are selected from at least one of a group consisting of alkyl mercury, alkenyl mercury, alkynyl mercury, aromatic mercury compounds, alkyl lead compounds, alkyl arsenic compounds and alkyl cadmium compounds.

43. A photosynthetic organism transformed with the vector of claim 38 which is useful for bioremediation of mercury and organomercurial compounds from contaminated water bodies.

44. A method of transforming a chloroplast of a selected photosynthetic organism to confer greater resistance to metal ions, said method comprising the steps of:

stably transforming the chloroplast of a photosynthetic organism with the vector of claim 38, growing the transformed photosynthetic organism under conditions which allow the expression of said expression cassette.

45. The method of claim 44, further comprising culturing said photosynthetic organism in a growth medium comprising a selector, and selecting transformed cells capable of growth in the presence of said selector.

46. The method of claim 45, further comprising regenerating a transgenic photosynthetic organism from said transformed cells.

47. A method of phytoremediation of mercury and organomercurials in bodies of contaminated water, said method comprising the steps of:

treating water contaminated with mercury and organomercurials with the transgenic photosynthetic organism of claim 42 before releasing the water into the environment.

48. The photosynthetic organism of claim 43, wherein said photosynthetic organism is either a green algae or a cyanobacteria.

49. The photosynthetic organism of claim 48, wherein the green algae is *Chlorella vulgaris.*

50. The photosynthetic organism of claim 48, wherein the cyanobacteria is *Synechocytis.*

51. A vector of claim 1 wherein a multi-gene operon codes for a protein.

52. A vector of claim 51, wherein the protein is a biopharmaceutical protein.

53. A vector of claim 52, wherein the biopharmaceutical protein is a monoclonal antibody.

54. A vector of claim 53, wherein the protein is produced in the same stoichiometric ratio.

55. A vector of claim 4, wherein said another gene of the operon is selected from the group of cholesterol oxidase, alpha-amylase inhibitors, and proteinase inhibitors.

56. The vector of any one of claims 1-13, 21-27, 37-42, 51-55, wherein the promoter is a one functional in green or non-green plastids.

57. The promoter of claim 56, wherein said promoter is selected from the group of psbA, accD, or 16srRNA promoters.

58. The biosynthetic pathway of claim 1, wherein said biosynthetic pathway result in the production of compounds such as amino acids, fatty acids, carbohydrates, polymers, vitamins, antibiotics and dyes.

59. A vector of claim 8, where the protein is human serum albumin.

60. A vector of claim 1, which further comprises flanking each side of the expression cassette, flanking DNA sequences which are homologous to a DNA sequence inclusive of a spacer sequence of the target chloroplast genome, which sequence is conserved in the chloroplast genome of different plant species, whereby stable integration of the heterologous coding sequence into the chloroplast genome of the target plant is facilitated through homologous recombination of the flanking sequences with the homologous sequences in the target chloroplast genome.